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AU - Neystat M; Rzhetskaya M; Chaudhari S; Yarygina O; Kholodilov N; Burke R E

AUAF- Columbia University, New York, NY, USA

CONF- 30th Annual Meeting of the Society of Neuroscience; New Orleans, LA, USA; November 04-09, 2000

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RN - 216864-07-2 (alpha-synuclein)

XNPL- 0190-5295-26-1-0

AB - Human alpha-synuclein was first identified as a component of Alzheimer senile plaques (Ueda et al, 1993). It has also been implicated in the pathogenesis of Parkinson's disease (PD). Two mutations (A53T and A30P) have been identified in some familial cases, and, in sporadic cases, alpha-synuclein has been identified in Lewy bodies. In spite of its importance to neurodegenerative disease, little is known of its normal function. In order to identify interacting protein partners with rat alpha-synuclein in brain we have performed a yeast two-hybrid screening analysis. A full length coding sequence for rat alpha-synuclein 1 (Kholodilov et al, 1997, AF007758) was expressed in the pGBKT7 DNA-BD vector of the Clontech Matchmaker system, and used to screen an adult rat whole brain cDNA library expressed in pACT2. Two candidate sequences have been identified. The first, 125, was represented by four clones; the second, 86, by only one. The sequences of both candidates were extended by 5' RACE. The 1036bp sequence of 125 contains an ORF for a 123aa protein; the 1124bp sequence of 86 for a 268aa protein. Neither has known homologues in GenBank. Northern analysis of immature rat substantia nigra (SN) (in which alpha-synuclein is abundantly expressed) demonstrates a single transcript of approximately 3.0 kb for 125, and of 1.1-1.4 kb for 86. In situ hybridization analysis confirms that both transcripts are expressed in SN pars compacta. To analyze regions of interaction between these candidate protein products and synuclein, two-hybrid interactions with sequences for aa 1-65, 60-95, and 88-140 of alpha-synuclein were assessed. The 125 protein interacted with aa 60-95 and 88-140, but not 1-65. The 86 protein interacted with all regions of alpha-synuclein. MS 26836, MS 38370, PDF, Lowenstein, Smart Foundations.

AUC - USA

AW - ** Methods and Equipment **

two-hybrid screen, analytical method

- ** Miscellaneous Descriptors **

Meeting Abstract

IW - ** Major Concepts **

Biochemistry and Molecular Biophysics; Nervous System (Neural

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